Propagation of human embryonic and induced pluripotent stem cells in an indirect co-culture system.

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Public Summary:
We developed a useful culture method in which human ES cells are cultured on a porous membrane separated from feeder layer cells.

Scientific Abstract:
We have developed and validated a microporous poly(ethylene terephthalate) membrane-based indirect co-culture system for human pluripotent stem cell (hPSC) propagation, which allows real-time conditioning of the culture medium with human fibroblasts while maintaining the complete separation of the two cell types. The propagation and pluripotent characteristics of a human embryonic stem cell (hESC) line and a human induced pluripotent stem cell (hiPSC) line were studied in prolonged culture in this system. We report that hPSCs cultured on membranes by indirect co-culture with fibroblasts were indistinguishable by multiple criteria from hPSCs cultured directly on a fibroblast feeder layer. Thus this co-culture system is a significant advance in hPSC culture methods, providing a facile stem cell expansion system with continuous medium conditioning while preventing mixing of hPSCs and feeder cells. This membrane culture method will enable testing of novel feeder cells and differentiation studies using co-culture with other cell types, and will simplify stepwise changes in culture conditions for staged differentiation protocols.